Assembly of root-associated microbiomes of typical rice cultivars in response to lindane pollution

Jiayin Feng[a,b], Yan Xu[a,b], Bin Ma[a,b], Caixian Tang[a,c], Philip C. Brookes[a,b], Yan He[a,b,*], Jianming Xu[a,b]

[a] Institute of Soil and Water Resources and Environmental Science, College of Environmental and Resource Sciences, Zhejiang University, Hangzhou 310058, China
[b] Zhejiang Provincial Key Laboratory of Agricultural Resources and Environment, Hangzhou 310058, China
[c] Department of Animal, Plant and Soil Sciences, La Trobe University, Bundoora, Victoria 3086, Australia

ABSTRACT

Organochlorine pesticides have been extensively used for many years to prevent insect diseases of rice (*Oryza sativa* L.), but little is known about their residual impacts on the underground micro-ecology in anaerobic environment. In this glasshouse study, we characterized the lindane effects on the assembly of root-associated microbiomes of commonly used indica, japonica and hybrid rice cultivars, and their feedback in turn, in modifying lindane anaerobic dissipation during 60 days' rice production. The results showed that rice growth inhibited the anaerobic dissipation of lindane, but was not affected apparently by lindane at initial spiked concentration of 4.62 and $18.54 \, \mathrm{mg \, kg^{-1}}$ soil. Suppressed removal of lindane in rice planted treatments as compared with that in unplanted control was likely due to inhibited reductive dechlorination induced by a comprehensive effect of radial O_2 secretion of rice root and co-occurring Fe(III) reduction that consumed electron competitively in rice rhizosphere. However, the hybrid cultivar exhibited a less suppression than the conventional cultivars in high polluted soils. Bacteria was more sensitively responded to lindane pollution than fungal taxa, and Actinobacteria, Chloroflexi, Verrucomicrobia and Proteobacteria were the main different phyla between hybrid and conventional cultivars, with a more stable community structure exhibited in the hybrid rice under lindane stress. Our study highlights the assembly and variation of root-associated microbiomes in responses of lindane pollution, and suggests that hybrid rice cultivar might be most competent for cultivation in paddy fields polluted by lindane and other organochlorine pesticides, especially in the area with high residual levels.

1. Introduction

Organochlorine pesticides (OCPs) were extensively used in the last century during agricultural production all over the world. Due to the stable aromatic ring structure and high chlorine content, they were extremely persistent to degrade, and still frequently detectable in most of farmland soils in China although they have already been banned from application up to almost 30 years (Rani et al., 2017; Salam et al., 2017; Xu et al., 2018; Zhu et al., 2018). For instance, as one of the oldest synthetic broad spectrum OCPs with insecticidal activity, lindane (γ-hexachlorocyclohexane, γ-HCH) was detected even up to > 20 $\mathrm{mg \, kg^{-1}}$ in soils, because of its low aqueous solubility and soil-sorption coefficient ($Koc = 1100 \, \mathrm{cm^3 \, g^{-1}}$) (Phillips et al., 2005; Feng et al., 2011). Therefore, it was widely acknowledged as a recalcitrant environmental issue, and crops were very likely grown under stress of OCPs.

Reductive dechlorination is the most important degradation processes for OCPs removal under waterlogged anaerobic soil environments (Xu et al., 2018). Such degradation process is an anaerobic microbe-mediated electron accepting process and driven by electrons flow from electron donors (such as carbon substrates, reducing minerals and hydrogen), during which OCPs act as an electron acceptor (Xu et al., 2015; Xue et al., 2017; Zhu et al., 2018). Previous studies have verified the reductive dechlorination usually coupled with natural soil redox processes, and other co-occurring electron acceptors in soil, such as Fe (III), may complete with OCPs for electrons to reduce under an electron limited condition, thereby suppressing the dechlorinated dissipation of OCPs (Xu et al., 2015; Lin et al., 2018).

* Corresponding author at: Institute of Soil and Water Resources and Environmental Science, College of Environmental and Resource Sciences, Zhejiang University, Hangzhou 310058, China.
E-mail address: yhe2006@zju.edu.cn (Y. He).

Rice (*Oryza sativa* L.) is one of the most important crops, feeding over half of the world's population (Zhang et al., 2019). The main cultivated rice, including indica, japonica and hybrid cultivars, are widely reported differently at root morphology, secretion release, radial oxygen loss (ROL) and genomic component (Wang et al., 2011; Bailey-Serres and Brinton, 2012; Zhao et al., 2018; Zhang et al., 2019), causing differential effects on rice yield, nutrient utilization and adversity stress resistance. As a hygrophyte, soluble organic carbon acids and O_2 secreted by rice roots might alter the redox and electron supply status of anaerobic environment, further influence the electron flow in soil complex reductive processes. To date, however, the underground micro-ecological responses to stress of OCPs during rice cultivation (especially cultivation of difference rice cultivars), and their feedback in modifying the anaerobic dissipation of OCPs during rice production, are not yet disclosed by on-going researches. This warrants relevant follow-up study.

In natural, plants could alter the root-associated environments through their impact on soil chemical properties and indigenous microorganisms resulted from the shaping role of root exudates (Mitter et al., 2017; Santos-Medellín et al., 2017). Recent studies have provided new insights into the assembly and variation of plant microbiomes by distinguishing different rhizo-compartments (bulk soils, rhizosphere, rhizoplane and endosphere) and suggested a selective effect of host plants on the establishment of root-associated communities (Edwards et al., 2015; Santos-Medellín et al., 2017). Microbes, including bacteria and fungi, inhabit the rhizosphere as a result of their functional traits, which might be directly or synergistically beneficial to plant nutrient acquisition and stresses resistance (Lu et al., 2018).

In this study, we used high-throughput sequencing of partial 16S rRNA gene and internal transcribed spacer region (ITS) amplicons to reveal the underground micro-ecological responses to lindane pollution during rice cultivation, with the special aims to disclose rice cultivar-dependent assembly of root-associated microbiomes and O_2 secretion of rice under lindane stress, and thereby their feedback in modifying lindane anaerobic dissipation during rice production. It was hypothesized that 1) contrasting rice cultivars (including hybrid, japonica and indica) may differ in root secretion and radial O_2 loss, resulting in cultivar-dependent modification for flooded soil environment; and 2) reductive dechlorination is the main pathway for removal of soil residual lindane during rice production, and this process might be inhibited by natural soil redox processes that can consume electron competitively.

2. Material and methods

2.1. Soil

Soil samples were collected from the rice field in Jiaxing, Zhejiang province of China (30° 50′8.74″ N, 120° 43′3.68″ E). The farmland is a representative area for rice cultivation in Yangtze River delta of China and harvest twice per year. Surface soil was collected using shovels to gather down to a depth of approximately 20 cm by the "S" sampling strategy in a 20 m × 20 m field. All soils were transported back to the greenhouse, spread evenly, air-dried in a ventilated square, passed through a 2-mm sieve and thoroughly mixed prior to the batch experiments. The basic physicochemical properties of the soil were as follows: pH (soil:water = 1:2.5) 6.76; organic matter, 54.4 g kg^{-1}; clay, 36.3%, and silt, 45.3%. Two pollution levels of lindane with low and high doses (5 and 20 mg kg^{-1}, respectively) were designed as our previous studies (He et al., 2005, 2015). The initial spiked concentrations were detected as 4.6 and 18.5 mg kg^{-1}, respectively.

2.2. Greenhouse experiment

The experiment was a completely factorial randomized block design, and consisted of four cultivation treatments and three pollution levels in three replicates. The four cultivation treatments were 1) the

control without plants (Control), and three contrasting cultivars of rice (*Oryza sativa* L.) (2) subsp. japonica "Xiushui 519" (XS), (3) subsp. indica "Huanghuazhan" (HHZ), and (4) hybrid "Yongyou12" (YY). The three lindane levels were "0", "5" and "20" (mg kg^{-1}). All the cultivars have been widely grown in eastern China, with obvious differences in plant physiological traits (He et al., 2015; Wei et al., 2017; Chu et al., 2018).

Uniform seeds were surface-sterilized in 70% ethanol for 30 s and 10% NaClO for 10 min, followed by washing with deionized water twice. The seed were then germinated on wet gauzes in the dark at 25 °C under sterile conditions for 24 h. The germinated seeds were transferred, and seedlings were cultivated in the Hoagland nutrient solution for 2 weeks. Three uniform seedlings were then selected and transplanted into pots (with 15 cm in height and 10 cm in diameter) with 1.8 kg lindane spiked soils. After tillering stage, two stems were removed, with only the most representative one left in each pot. The plants were then grown for 60 days in a naturally-lit greenhouse with average day/night temperatures of 28/20 °C, and day/night humidity of 70%/85%. Each pot was watered every other day with around 200-ml sterilized deionized water to control the flooded condition. After 60 days' growth, the plant tissues were collected and divided into two parts: one was used for contaminant analysis, and the other for root-associated compartments sampling.

2.3. Sampling of bulk soils, rhizosphere and endosphere

The method for sampling bulk soils, rhizosphere and endosphere compartments followed the protocols described in Bulgarelli et al. (2012) and Edwards et al. (2015) with some modifications. Briefly, loosely bound soil was manually removed, leaving approximately 1 mm soil firmly attached to the roots. The roots were then placed in a sterile 50-ml falcon tube containing 30 ml of sterile phosphate-buffered solution (PBS) (with pH around 7.4) and vortexed at 3000 rpm for 15 s, and the turbid solution was filtered through a 100-μm aseptic nylon mesh strainer into a new 50-ml tube to remove root fragments and large sediments, followed by centrifuging for 2 min at 12,000 × *g*. The supernatant was discarded, and the soil washed from the roots was defined as rhizosphere soil, frozen in liquid nitrogen, and stored at −80 °C. The washed roots were transferred to a falcon tube with 30 ml PBS and sonicated for 30 s at 50–60 Hz with 3 cycles to strip the soil firmly adhering to the root surface, and then transferred into a 50-ml tube with 30 ml PBS and washed twice. Finally, the roots were stored as an endosphere samples at −80 °C until DNA extraction.

Bulk soil samples were collected from unplanted pots, other than from the pot with growing plants, as the impact distance of O_2 secretion of rice roots was usually beyond rhizosphere area what we commonly defined. This sampling method could completely eliminate any possible effects from rice roots. After collection, bulk soil samples were mix thoroughly and stored at −80 °C.

2.4. Chemical analysis of soil and plants

Lindane (99% pure) was purchased from Sigma-Aldrich Co. (St. Louis, USA). The extractants (> 99.9% purity, HPLC), including n-hexane and acetone, were obtained from Tedia Co., Inc. (Fairfield, USA) and ANPEL Laboratory Technologies Inc. (Shanghai, China). Other analytical grade chemicals were obtained from Sinopharm Chemical Reagent Co., Ltd., Shanghai, China. Anhydrous sodium sulfate was muffle furnace-dried at 750 °C for 4 h before use.

The concentrations of lindane in soil, roots and shoots were extracted by a method based on accelerated solvent extraction (ASE) with Dionex ASE 350 (Sunnyvale, USA), and subsequent solid phase enriched, followed by GC–MS analysis according to Popp et al. (1997) with some modification. Detailed procedures of extraction and detection can be found in the SI.

Soil pH was determined in a suspension of 1:2.5 soil/water ratio (*w/*

Table 1
The height and biomass of different rice cultivars, and their accumulation amount for lindane.

Cultivars[a]	Pollution dose[b]	Root			Shoot		
		Length	Biomass	Lindane	Height	Biomass	Lindane
	(mg kg^{-1})	(cm)	(g FW plant^{-1})	(μg kg^{-1})	(cm)	(g FW plant^{-1})	(μg kg^{-1})
XS	0	19.6 ± 1.2cd[c]	5.04 ± 0.65bc	nd[d]	69.7 ± 0.9e	24.7 ± 2.81 cd	nd
	5	18.2 ± 2.4d	5.00 ± 0.37bc	59.1 ± 13.5b	66.4 ± 1.4f	25.2 ± 0.75 cd	16.5 ± 6.81ab
	20	19.1 ± 1.5 cd	4.12 ± 0.19c	164.0 ± 40.0a	68.0 ± 1.5ef	20.8 ± 1.05d	31.2 ± 10.1a
HHZ	0	23.3 ± 1.6a	6.75 ± 0.58a	nd	83.2 ± 1.1bcd	34.4 ± 2.80ab	nd
	5	22.5 ± 1.3ab	6.09 ± 0.36ab	64.7 ± 49.6b	87.5 ± 1.2ab	36.0 ± 5.50ab	nd
	20	23.1 ± 1.6a	4.12 ± 0.60c	137.4 ± 43.2a	88.1 ± 4.9a	32.2 ± 0.43bc	26.7 ± 15.4a
YY	0	20.2 ± 1.8bcd	5.97 ± 0.29ab	nd	80.5 ± 1.9d	42.0 ± 3.65a	nd
	5	21.3 ± 1.0abc	6.09 ± 0.71ab	72.3 ± 16.1b	81.3 ± 2.1 cd	40.1 ± 1.89a	6.4 ± 2.62b
	20	21.5 ± 0.7abc	6.75 ± 1.25a	167.3 ± 28.8a	85.7 ± 0.8abc	37.2 ± 7.20ab	22.3 ± 14.7ab

[a] Abbreviations: XS, japonica cultivar XS519; HHZ, indica cultivar HHZ; YY, hybrid cultivar YY12.
[b] Lindane spiked concentration (mg kg^{-1}).
[c] Values are means ± standard errors of three replications; values within a column followed by a common letter are not significantly different ($P < 0.05$).
[d] nd: not detected.

v) with a pH meter (S975 SevenExcellence, MettlerToledo, Switzerland). Concentrations of sulfate, dissolved organic carbon (DOC) and dissolved total nitrogen (DTN) were extracted with Milli-Q water, and the supernatant was measured using ion chromatography (Dionex ICS-2000, United States), and automated total organic carbon analyzer (Multi N/C 3100, Analytik Jena AG, Jena, Germany), respectively (Xu et al., 2015; Dai et al., 2016). The concentration of Fe^{2+} was measured using the 1,10-phenanthroline colorimetric method at 530 nm on a UV–Vis spectrophotometer (Xu et al., 2015). The root radial oxygen loss (ROL) was determined using a miniaturised Clark-type oxygen micro-electrode system (OXY25, ø020–30 μm, Unisense, Aarhus, Denmark) for 10-h sequential in situ detection, which has been described in Li and Wang (2013). Fresh biomass of shoots and roots were recorded.

2.5. DNA extraction, amplicon amplification and sequencing analysis

The total genomic DNA of each sample was extracted using FastDNA SPIN kit (Mpbio, United States) for soils and roots according to the manufacturer's instructions. The quantity and quality of extracted DNA were checked photometrically using a NanoDrop ND-1000 UVeVis spectrophotometer (NanoDrop Technologies, Wilmington, DE, United States). PCR amplification of bacterial 16S rRNA genes (V4 region) and fungal ITS (ITS1 region) were performed using the primer 515F (5′-GTGCCAGCMGCCGCGGTAA-3′) and 806R (5′-GGACTACHVGGGT-WTCTAAT-3′), and ITS5F (5′-GGAAGTAAAAGTCGTAACAAGG-3′) and ITD1R (5′-GCTGCGTTCTTCATCGATGC-3′), respectively. The PCR amplicons were purified with Agencourt AMPure Beads (Beckman Coulter, Indianapolis, IN) and quantified using the PicoGreen dsDNA Assay Kit (Invitrogen, Carlsbad, CA, USA). After the individual quantification step, amplicons were pooled in equal amounts, and paired-end 2 × 300 bp sequencing was performed using the Illumina MiSeq platform with MiSeq Reagent Kit v3 at Shanghai Personal Biotechnology Co., Ltd. (Shanghai, China).

The Quantitative Insights Into Microbial Ecology (QIIME, v1.8.0) pipeline was employed to process the sequencing data, as previously described (Caporaso et al., 2010). The high-quality sequences were clustered into operational taxonomic units (OTUs) at 97% sequence identity by UCLUST. OTU taxonomic classification was conducted by BLAST searching the representative sequences set against the SILVA and ITS Database. After trimming and filtration, the quality sequences were clustered into 5134 and 2462 operational taxonomic units (OTUs) with 97% similarity for bacteria and fungi, respectively. All sequences were submitted to the NCBI Sequence Read Archive (SRA) database with accession number SRP165249.

2.6. Statistical analysis

Analysis of lindane dissipation and microbial variation was based on ANOVA comparison with LSD test at $P < 0.05$ using SPSS 20.0 statistical software (IBM, Armonk, IL, United States). Principal coordinate analysis (PCoA) and permutational multivariate analyses of variance (PERMANOVA) by Bray-Curtis distance matrix were performed to assess the differences in community composition (Liu et al., 2015). The P-value was corrected for multiple testing using the method of Benjamini and Hochberg (1995), and a false discovery rate (FDR) was used as a threshold to denote statistical significance (Peperanney et al., 2016). Statistical analyses of differentially abundant OTUs were performed using the edgeR package (Robinson et al., 2010). All graphs were produced with ggplot2 package in R (v 3.4.0) and OriginPro 2017.

3. Results

3.1. Plant growth and lindane concentration in rice tissues and soils

After 60 days of growth, no visible symptoms of lindane toxicity were observed for all cultivars of rice. The root length, shoot height and biomass of roots and shoots presented no consistent difference between polluted and unpolluted treatments (Table 1). The hybrid YY showed the maximum biomass in both roots and shoots (6.75 g and 41.95 g respectively), and indica HHZ had the highest while japonica XS the lowest shoot height and root length (Table 1).

The concentrations of lindane in root tissues among the cultivars were in the range of 59.1–72.3 μg kg^{-1} and 137.4–167.3 μg kg^{-1} at lindane doses of 5 and 20 mg kg^{-1}, respectively (Table 1). Shoot tissues accumulated much lower lindane than the root, mostly only detected at 20 mg kg^{-1} of lindane dose with a range of 26.7–32.3 μg kg^{-1} (Table 1). Additionally, the unplanted control resulted in the lowest concentration of residual lindane in soils, with 0.09 and 0.25 mg kg^{-1} at 5 and 20 mg kg^{-1} of lindane, respectively; while rice growth significantly inhibited lindane dissipation in soils in highly polluted soil, with a residual concentration ranging from 0.30 to 0.40 mg kg^{-1} ($P < 0.05$). Among the three cultivars, lindane concentration in rhizosphere soils of hybrid YY was significantly lower than those of conventional indica HHZ and japonica XS at highly polluted treatments ($P < 0.05$) (Fig. 1A).

The rates of root radial oxygenation loss (ROL) differed among the cultivars, ranging from 118 to 135 μmol L^{-1} The hybrid YY had the lowest O_2 content with an average of 120 μmol L^{-1}, while the rate of ROL of XS and HHZ were 129 and 133 μmol L^{-1}, respectively (Fig. 1B). Additionally, the concentrations of DTN, Fe^{2+} and SO_4^{2-}, and soil pH
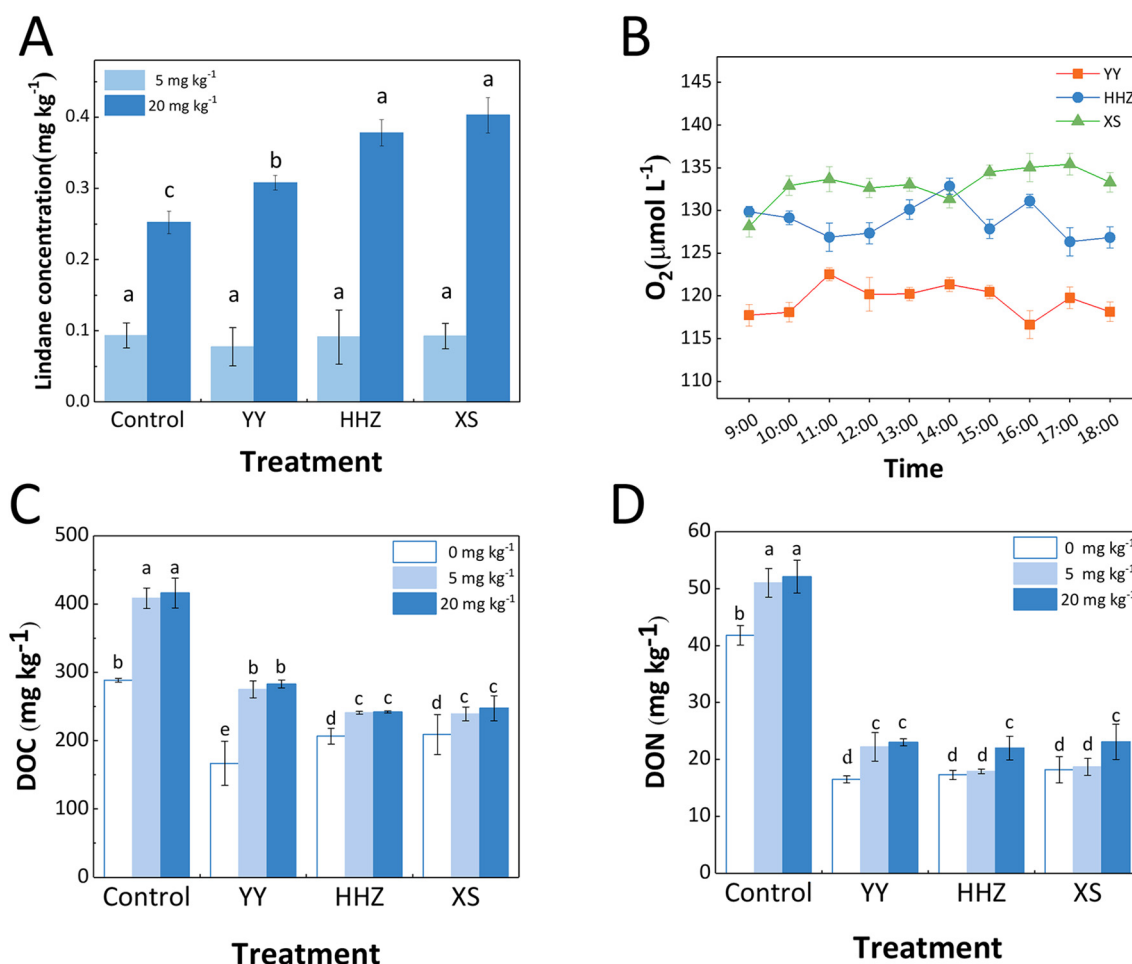
Fig. 1. The concentration of residual lindane in rhizosphere soils after 60-day growth of rice (A), the average radial oxygen loss (ROL) among different rice cultivars (B), and concentrations of dissolved organic carbon (DOC) (C) and dissolved total nitrogen (DTN) (D) among three rice cultivars at lindane doses of 0, 5 and 20 mg kg^{-1}. Bars are the standard error of means of three replicates. Different letters indicate significant differences among treatments at the $P < 0.05$ level. Abbreviations for treatments: Control: the control without rice; XS, japonica cultivar XS519; HHZ, indica cultivar HHZ; YY, hybrid cultivar YY12.

exhibited no obvious difference among the cultivars ($P > 0.05$), while soil DOC in polluted treatments was significantly higher with hybrid than with conventional rice ($P < 0.05$) (Table S1, Fig. 1C and D). Meanwhile, the polluted treatments generally had higher soil DOC concentration than non-polluted treatments ($P < 0.05$) (Fig. 1C and D).

3.2. Composition and diversity of microbial community

For bacteria community, Shannon index was used to measure the α-diversity indices. Samples from highly polluted soils had lower α-diversity than those from unpolluted soils in unplanted control, while lindane pollution affected the α-diversity in endosphere compartments of rice planted treatments ($P < 0.05$) (Fig. S1). There was no consistent difference, among rice cultivars, but Shannon index in the endosphere was significantly lower than that in the rhizosphere and bulk soils for both bacteria and fungi ($P < 0.05$) (Fig. S2).

There were notable differences in the proportions of bacteria (at the phylum level) and fungi (at the class level) between pollution dose and compartments among the treatments (Fig. S1). The bacterial community was dominated by Chloroflexi, Proteobacteria and Actinobacteria, along with the relative abundance of Acidobacteria decreased in both rhizosphere and endosphere, while Firmicutes increased in soils (bulk soils and rhizosphere). As for the fungal community, the phyla of Basidiomycota and Ascomycota account for over 70% of the reads. The relative abundance of *Agaricomycetes*, *Sordariomycetes* and

Tremellomycetes accounted for two thirds of the total abundance in the rhizosphere community, with *Sordariomycetes* decreased obviously from rhizosphere to endosphere. Nevertheless, the other two classes increased under YY and HHZ (Fig. S1D). There was no significant difference of these groups between pollution dose treatments.

Permutational multivariate analysis of variance (PERMANOVA) were used to investigate the dependence of variations in root-associated microbiomes to the effect of rice cultivars, pollution doses and compartments individually or in combination, based on Bray-Curtis distance (Table 2). For the bacterial community, the compartments comprise the largest source of the observed variation in the microbiome data (71.65%, $P < 0.001$), followed by cultivars (2.98%, $P < 0.05$) and pollution doses (2.49%, $P < 0.05$). In comparison, for the fungal community, the compartments and cultivars accounted for a larger proportion of variation (47.37%, $P < 0.001$ and 21.98%, $P < 0.001$ individually) than pollution doses (2.12%, $P < 0.05$) (Table 2). This result was confirmed by the PCoA analyses (Fig. 2).

3.3. Lindane stress on rice root-associated microbiomes

Fig. 3 illustrates the net proportional changes in relative abundances of bacterial and fungal communities responded to lindane pollution at the family level. There were 10 and 6 families of bacteria exhibited significant differences between polluted and unpolluted treatments in the rhizosphere and endosphere, respectively. The families of *Bacillaceae* and *Comamonadaceae* increased by 1.45% and 0.46% ($P < 0.05$)

Table 2
The environmental variables affecting the microbial community composition as revealed by PERMANOVA[a].

	Bacteria				Fungi			
	SS	Variation (%)[c]	F.Model	*P* value[d]	SS	Variation (%)	F.Model	*P* value
Cultivars	0.36	2.98	3.93	0.014*	6.74	47.37	56.2	0.001***
Pollution doses	0.30	2.49	3.29	0.025*	0.30	2.12	2.51	0.018*
Compartments	8.65	71.65	189.15	0.001***	3.13	21.98	52.14	0.001***
Cultivars × pollution doses[b]	0.35	2.90	1.91	0.09	0.48	3.38	2.00	0.033*
Cultivars × compartments	0.30	2.46	3.25	0.026*	0.84	5.88	6.98	0.001***
Pollution doses × compartments	0.20	1.70	2.24	0.073	0.20	1.39	1.65	0.122
Cultivars × pollution doses × compartments	0.26	2.18	1.44	0.208	0.38	2.70	1.60	0.087
Residuals	1.65	13.64			2.16	15.17		
Total	12.08				14.22			

[a] PERMANOVA, permutational multivariate analysis of variance; SS, sums of squares.
[b] "×" between the environmental variables indicates interactions between these variables.
[c] Variation was based on Bray-Curtis distances.
[d] *P* value based on PERMANOVA (999 permutations).
* $P < 0.05$.
*** $P < 0.001$.

with a significant enrichment in the rhizosphere, while the relative abundance of *Solibacteraceae*, *Gallionellaceae* and *Streptomycetaceae* decreased by 0.47%, 0.46% and 0.33% in polluted treatments compared to the unpolluted treatments. In the endosphere, the families of *Nocardioidaceae* and *Geobacteraceae* in the polluted treatments decreased by 0.27% and 0.19%, respectively, while *Roseiflexaceae* increased by 0.79%. As for the fungal community, only *Lasiosphaeriaceae* showed significant differences, decreasing by 0.06% in the polluted treatments.

The numbers of enriched OTUs in each compartment were higher in the polluted than unpolluted treatments (90, 99 and 433 OTUs versus 22, 21 and 360 OTUs in bulk soils, rhizosphere and endosphere, respectively) (Fig. 4A and B). Fungi community was dominant in differences among all soil compartments, while bacteria led to the difference primarily in the endosphere (only 2 OTUs of bacteria in bulk soil and none in rhizosphere) (Fig. 4C and D). Proteobacteria, Actinobacteria, Chloroflexi and Firmicutes, Basidiomycota and Ascomycota accounted for the vast majority of enriched OTUs in each compartment (Fig. 4C and D) and the amount of OTUs belonging to these phyla were higher in the polluted than unpolluted treatments.

3.4. Cultivar difference in microbial responses to lindane addition

Given that the lindane dissipation rate in highly polluted soils of YY treatment was significantly higher than that in XS and HHZ, conventional XS and HHZ were grouped together for exploring the variation of microbial communities. The conventional cultivars had higher cumulative relative abundances of lindane-responsive OTUs than hybrid rice in both rhizosphere and endosphere. Within a given cultivar, the response of OTUs (both enriched and depleted) was greater in the endosphere than in the rhizosphere (Fig. 5A). Compared to the hybrid rice, 27 OTUs increased, and 60 reduced the relative abundance in conventional cultivars under lindane stress (Fig. 5B). There were noticeable differences in these OTUs between hybrid and conventional cultivars, in both rhizosphere and endosphere. Actinobacteria, Chloroflexi and Acidobacteria were mostly enriched in the hybrid rice, while Verrucomicrobia and Bacteroidetes were the main enriched phyla in the conventional cultivars. However, in the endosphere, Proteobacteria, Firmicutes and Fibrobacteres were enriched in the hybrid rice, but only Bacteroidetes enriched in the conventional cultivars (Fig. 5C).

There was a small difference (only 1 OTU enriched in polluted treatment of hybrid rice) in the fungal community between polluted and unpolluted treatments (Fig. S3). The number of differential OTUs was lower in the conventional than hybrid cultivar under lindane stress. *Scleroderma* and *Bullera* were relatively enriched under the hybrid rice compared to the conventional cultivars (Fig. S3).

4. Discussion

4.1. Effects of environmental variables on the dissipation of lindane

The addition of lindane at 5 and $20 \, mg \, kg^{-1}$ did not affect rice morphology on plant biomass, root length, and shoot height after 60 days (Table 1). Consistently, Kidd et al. (2008) indicated that *Cytisus striatus* and *Holcus lanatus* could grow with no visible signs of toxicity in
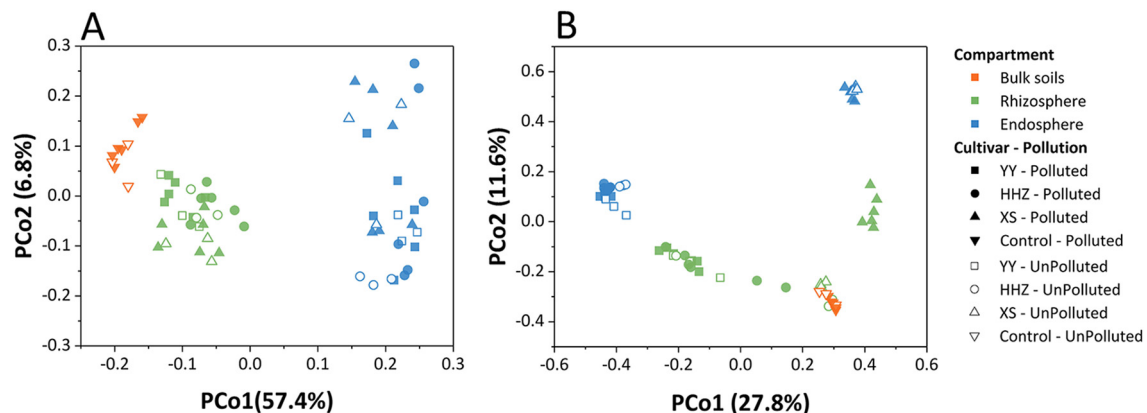
Fig. 2. PCoA analysis of bacterial (A) and fungal (B) communities among different treatments. Abbreviations are as Fig. 1.
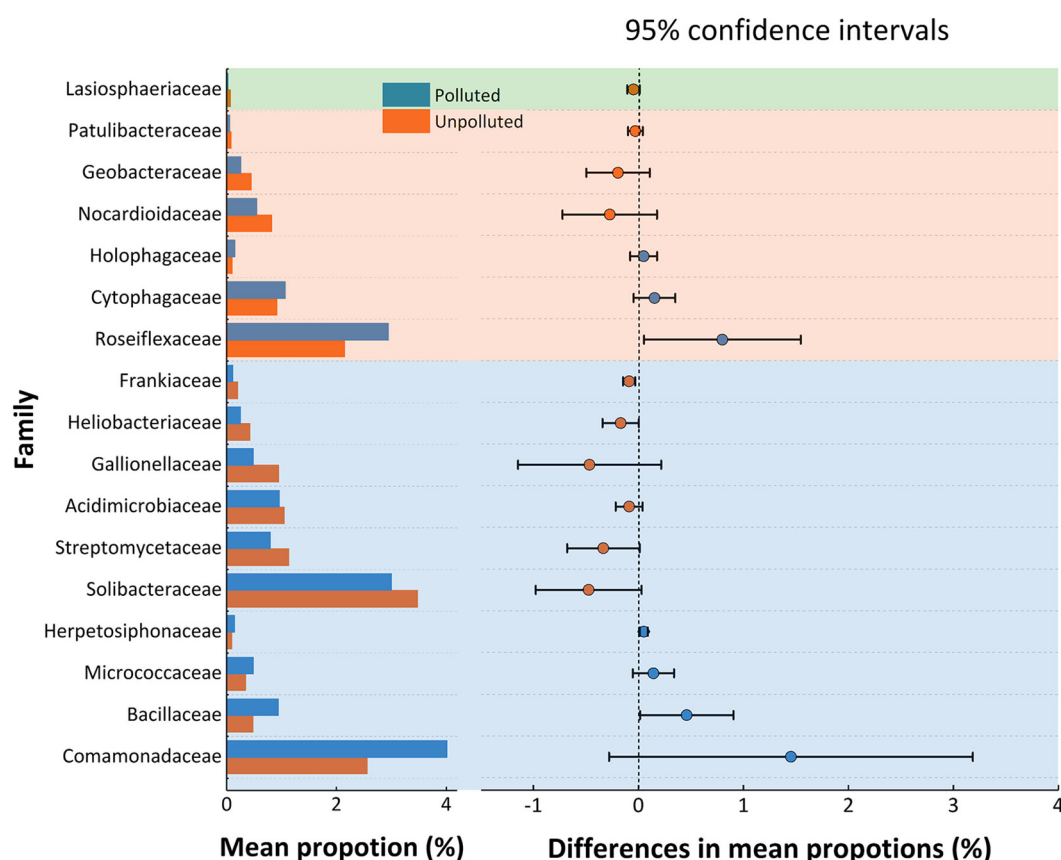
Fig. 3. Variation analysis of microbial community in rhizosphere and endosphere compartments at the family level in lindane-polluted treatments as comparison of that in unpolluted treatments. Blue bars and points stand for polluted treatments, and orange bars and points stand for unpolluted treatments. The blue background represents bacteria in the rhizosphere compartment, the orange background represents bacteria in the endosphere compartment, and the green background represents fungal community. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

soils polluted with up to $100\,\text{mg HCH kg}^{-1}$, and unlikely absorb and translocate HCH from soils to plant tissues. However, some other studies showed a negative influence of lindane on *Avena sativa*, *Solanum nigrum*, *Sesamum indicum* and *Vicia sativa*, with the highest accumulation of lindane around 20% in plant roots (Calvelo Pereira et al., 2006; Kidd et al., 2008; Becerra-Castro et al., 2013). In our study, rice cultivars did not accumulate much lindane, with the average phyto-accumulation of 1.4% and 0.8% at low and high doses, respectively (Table 1). This might be due to its higher hydrophobicity resulting in the slow movement of lindane into the roots and shoots via apoplastic and symplastic pathways (Su and Zhu, 2007), and the fast dissipation rate of lindane in anaerobic environment caused by soil-microbe interactions (Phillips et al., 2005). Under such a circumstance, rice would be a suitable crop for growing in lindane-polluted soils.

To date, many previous reports have verified the ability of plants to facilitate the dissipation of organochlorine pollutants, but most of them were relevant to dryland species. For example, maize, tomato and sunflower plants were reported capable of coping with high levels of organochlorine pesticide pollution, including lindane and endosulfan, and as a result of this, it was successfully applied for remediation of xenobiotics (Calvelo Pereira et al., 2006; Abhilash et al., 2013; Álvarez et al., 2015; Mitton et al., 2016). In contrast to the above studies, we found that rice growth indeed decreased the dissipation of lindane in their rhizosphere, with the concentration of lindane lower in the control than cultivation treatments. In our study, planted treatments had much higher Fe^{2+} contents than unplanted control, indicating a more active Fe(III) reduction under soil environment with rice growth (Table S1). Our previous results had showed that Fe(III) can be served as an alternative terminal electron acceptor to compete for electrons with OCPs

and split electron transferred from dechlorination to Fe(III) reduction during anaerobic redox processes in flooded soils (Xu et al., 2015; Lin et al., 2018; Zhu et al., 2018). The root exudates released by rice in planted treatment provided additional low-molecular-weight organic compounds as carbon sources as well as electron donors to improve microbial-initiated anaerobic respiration (Heidelberg et al., 2002; Hayat et al., 2011). This would likely promote the occurrence of more competitive redox processes such as Fe(III) reduction and thereby suppressing the reductive dechlorinated dissipation of lindane in rice planted condition. Especially, with a higher concentration of soil DOC, the hybrid rice likely provided more labile nutrients for rhizospheric microbes, thereby alleviated suppression from soil indigenous redox processes on the dechlorinated dissipation of lindane in their rhizosphere (Fig. 1C). Additionally, many wetland plants are capable of secreting radial oxygen along the root, which changes their rhizosphere environment and 'protects' roots against soil-derived toxins (Colmer, 2003; Wang et al., 2011). For example, rice cultivars with greater ability of ROL played an important role in inhibiting the accumulation and transfer of As and Cd to above-ground tissues (Mei et al., 2009; Wang et al., 2011); and an indica genotype with greater ROL formed iron plaque to sequester Sb in the root comparing with hybrid genotypes (Zhang et al., 2017). Given that lindane degradation was much faster in anaerobic condition than aerobic environment (Phillips et al., 2005), it was speculated that the secretion of O_2 might change the anaerobic to micro-aerobic environment during rice growth, thus inhibiting the anaerobic dissipation of lindane in the rhizosphere. This possibility was supported by the higher ROL for XS and HHZ than for YY (Fig. 1B). Thus ROL might be another important factor responsible for the suppressed dissipation of lindane in rice rhizosphere.
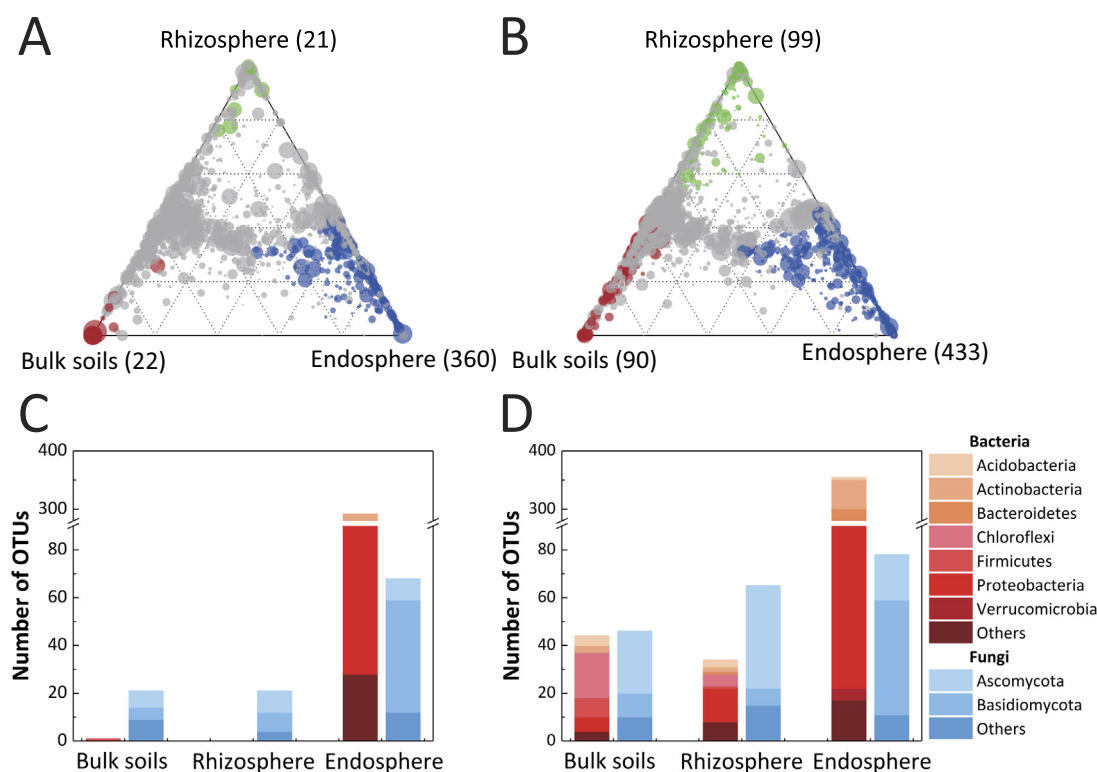
Fig. 4. OTU enrichment of bacterial and fungal communities bulk soils, rhizosphere and endosphere after 60-d plant growth. Ternary plots represent proportional contribution of OTUs (total relative abundance of all treatments > 0.1%) enriched in one compartment across unpolluted (A) and polluted (B) treatments. Bulk soil OTUs (red), rhizosphere OTUs (green), endosphere OTUs (blue), and insignificant OTUs (grey). Each circle represents one OTU. The size of each circle reflects the relative abundance. The number in the bracket represents the amount of significant OTUs in each compartment. Histogram represents the enriched OTUs of bacteria and fungi in each compartment of unpolluted (C) and polluted (D) treatments. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Worthy mentioning, the decreased dissipation of lindane in the rice rhizosphere might give a preliminary evidence regarding the persistence of organochlorine pesticides, such as lindane (combined with its homogenous isomers) in paddy fields due to rice plantation (Rani et al., 2017; Salam et al., 2017). With this respect, hybrid rice would have less adverse impact on self-purification of lindane-polluted soils, and thus may be recommended for rice production of the paddy fields polluted with organochlorine pesticides such as lindane instead of conventional rice.

4.2. Lindane-induced differences in the assembly of root-associated microbiomes

This study showed that no significant difference was exhibited for the microbial α-diversity of both bacterial and fungal communities with lindane pollution as comparison with unpolluted treatments (Figs. S1 and S2), but the shift in the biotic environment induced by rice growth may indirectly influence community functioning (Bell et al., 2014).

Bacteria are generally considered the primary degraders in contaminated soils, and the extent of hydrocarbon or organochlorine pollutants in soils has been correlated with the activity of specific bacterial groups (Bell et al., 2014). In our study, lindane pollution increased the relative abundance of *Bacillaceae* and *Comamonadaceae* but decreased *Streptomycetaceae*, *Solibacteraceae* and *Gallionellaceae* (Fig. 3). As previously reported, the family of *Comamonadaceae* extensively participated in some reduction processes, and some members were capable of degrading PCB, PCP, and 2,4-dichlorophenol (2,4-DCP) (Dallinger and Horn, 2014; Lin et al., 2016; Yu et al., 2016). *Bacillaceae* had similar effects on aldrin, endosulfan, lindane and atrazine (López et al., 2005; Sagarkar et al., 2013; Ishag et al., 2017). In contrast, lindane addition suppressed some members of *Streptomycetaceae*, *Solibacteraceae* and

Gallionellaceae. Especially, the family *Streptomycetaceae* had shown a great potential for biodegradation of organochlorine pesticides (lindane and chlordane) (Cuozzo et al., 2009, 2012; Álvarez et al., 2012; Saez et al., 2014) and was used widely as soil inocula because of the active mycelial growth (Shelton et al., 1996). Nevertheless, there was no sufficient evidence proving its dominance and ability in an anaerobic environment.

Lindane addition slightly decreased the abundance of fungal families of *Lasiosphaeriaceae*, belonging to *Sordariomycetes*, but did not affect other two abundant classes *Agaricomycetes* and *Tremellomycetes* (Figs. 4 and S1). *Sordariomycetes* is one of the largest classes of Ascomycota including many important endophytes, saprobes and epiphytes taxa (Maharachchikumbura et al., 2016), but few studies have reported its resistance or degradation ability on pesticides. Meanwhile, Wick et al. (2010) suspected that some fungi may facilitate hydrocarbon bioremediation in soils through their hyphae in order to increase bacterial access to hydrophobic substrates. As such, the fungal community might be synergistic with degrading bacteria on the dissipation of lindane in rhizosphere soils.

This study clearly showed a considerable fraction of OTUs enriched in the endosphere as compared to the other two compartments (Fig. 4), it suggested that endosphere was more independent and exclusive, possessing microbiomes that differed greatly from those of the bulk soil and rhizosphere (Edwards et al., 2015; Chen et al., 2018). Meanwhile, lindane addition expanded the differences between spatially separable compartments (as 90 vs 22, 99 vs 21 and 433 vs 360 OTUs in bulk soils, rhizosphere and endosphere, respectively) (Fig. 4A and B). Similarly, Chen et al. (2018) found that *Hibiscus cannabinus* roots selected specific metal-tolerant and plant growth-promoting bacteria under heavy-metal stress and resulted in the greater differences between root-soil compartments. Therefore, the responses of root-associated microbiomes to
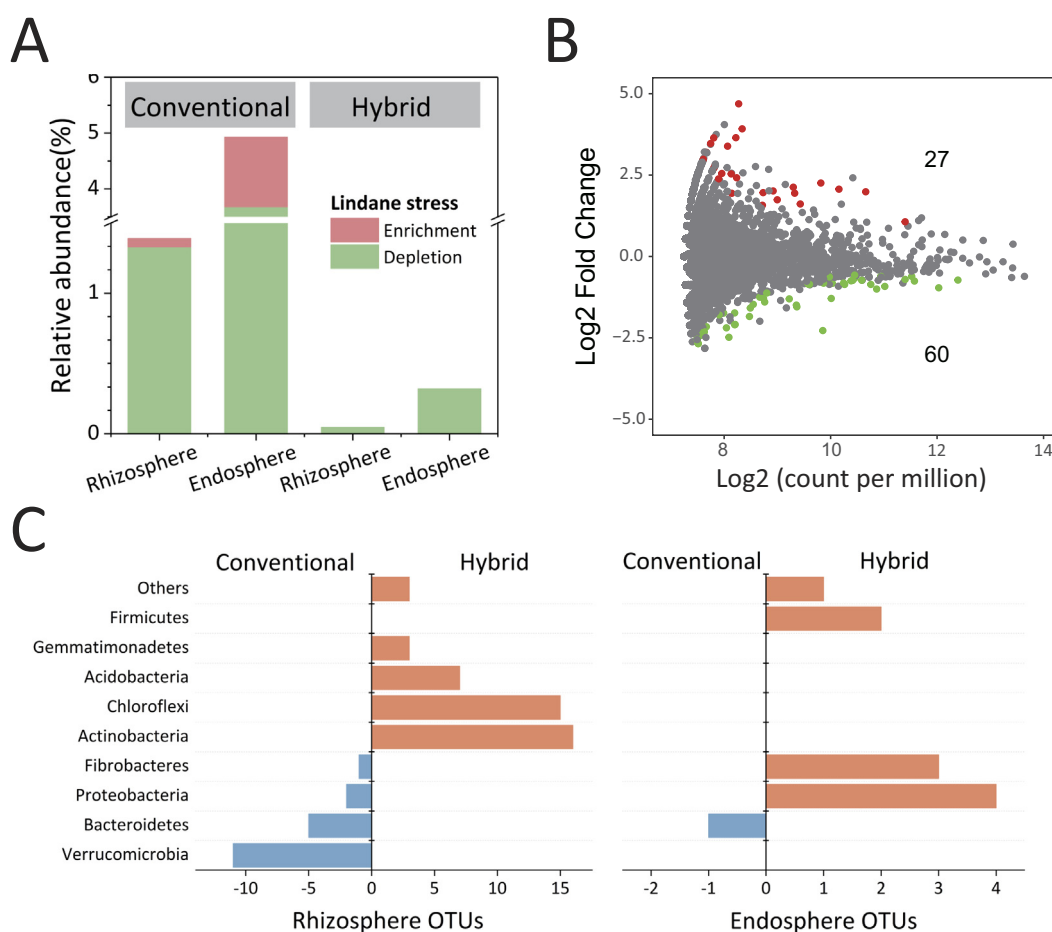
Fig. 5. Cultivar difference in bacterial OTUs in response to lindane pollution. (A) Cumulative relative abundances of lindane stress-responsive OTUs in conventional and hybrid rice microbial communities. (B) Volcano plots showing differential OTUs of conventional and hybrid cultivars. Taking hybrid rice as the control, red points and corresponding number indicated significant enrichment of OTUs in conventional cultivars, green points and corresponding number indicated depletion of OTUs in conventional cultivar ($P < 0.05$). Each point represents an individual OTU. (C) The enriched rhizosphere and endosphere OTUs of bacteria among hybrid and conventional rice. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

lindane addition differed in different compartments.

4.3. Cultivar difference in the assembly of root-associated microbiomes under lindane stress

Rice cultivar exhibited a significant effect on root-associated microbial communities (Table 2 and Fig. 2). Similarly, Shenton et al. (2016) indicated that root-associated bacterial communities showed small but significant differences between wild and cultivated rice. Edwards et al. (2015) contrasted 6 rice genotypes in greenhouse experiments including the japonica varieties, indica varieties and African cultivated rice, and revealed a significant influence of rice genotypes on microbial communities, especially on the rhizosphere microbiome. It was also reported the microbial diversity of two japonica cultivars were similar to the indica cultivar IR50 (Edwards et al., 2015). Meanwhile, several studies have explored the variation of root-associate microbiomes by the groups of plant lineages, functional or phenotypic differences (Walters et al., 2018; Peiffer et al., 2013; Shenton et al., 2016). Given that the higher dissipation rate and lower accumulation of lindane found in the rhizosphere and endosphere of hybrid cultivar, it accounted for a small proportion of cumulative relative abundance within microbial communities (Fig. 5A). The results suggested hybrid rice had a more stable microbial community than conventional cultivars especially under lindane pollution.

Compared with conventional cultivars, hybrid rice enriched the relative abundance of Acidobacteria, Actinobacteria, Chloroflexi in the rhizosphere and Proteobacteria, Firmicutes, Fibrobacteres in the endosphere under lindane stress (Fig. 5C). Edwards et al. (2015) indicated that microbial colonization of rice roots was not a passive process and that plants had the ability to select for certain microbial consortia or that some microbes were better at filling the root colonizing niche. This confirmed our results that many sensitive members of above phyla had active response to lindane pollution and subsequently selected by hybrid rice roots to rebuild a more stable and stress-resistant microbial community structure (Figs. 3 and 5) (Dallinger and Horn, 2014; Lin et al., 2016; Sun et al., 2017).

In spite of the insignificant influence of fungi on lindane dissipation, hybrid rice exhibited a higher α-diversity and more OTUs enriched in rhizosphere and endosphere compared with conventional cultivars (Figs. S1 and S3). This revealed that fungi might make contribution to the more stable root-associate microbiomes of hybrid rice, thus playing synergistic effects with bacteria (Bell et al., 2014).

5. Conclusion

This study demonstrated that rice growth inhibited the dissipation of lindane with the hybrid rice having a less suppression than the conventional cultivars. Cultivar difference in radical oxygen loss and active soil redox processes in the rhizosphere might contribute to the differences in dissipation of lindane and the assembly of root-associate microbiomes. Fungal community was disproportionately sensitive to lindane pollution, but might have synergistic effects with bacteria.

Lindane addition promoted *Bacillaceae* and *Comamonadaceae* in the rhizosphere while microbial colonization of roots was selected by rice cultivars. This study highlights the cultivar variation in bacterial and fungal microbiomes in response to pollution of organochlorine pesticides, and suggests that hybrid rice might be the competent rice for paddy fields polluted by organochlorine pesticides such as lindane.

Declaration of Competing Interest

The authors declare no conflict of interest.

Acknowledgements

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envint.2019.104975.

References

Abhilash, P.C., Singh, B., Srivastava, P., Schaeffer, A., Singh, N., 2013. Remediation of lindane by Jatropha curcas L: utilization of multipurpose species for rhizoremediation. Biomass Bioenergy 51, 189–193. https://doi.org/10.1016/j.biombioe.2013.01.028.

Álvarez, A., Yañez, M.L., Benimeli, C.S., Amoroso, M.J., 2012. Maize plants (Zea mays) root exudates enhance lindane removal by native Streptomyces strains. Int. Biodeterior. Biodegradation 66, 14–18. https://doi.org/10.1016/j.ibiod.2011.10.001.

Álvarez, A., Benimeli, C.S., Saez, J.M., Giuliano, A., Amoroso, M.J., 2015. Lindane removal using Streptomyces strains and maize plants: a biological system for reducing pesticides in soils. Plant Soil 395, 401–413. https://doi.org/10.1007/s11104-015-2575-5.

Bailey-Serres, J., Brinton, L.E., 2012. Waterproofing crops: effective flooding survival strategies. Plant Physiol. 160 (4), 1698–1709. https://doi.org/10.2307/41812018.

Becerra-Castro, C., Kidd, P.S., Rodríguez-Garrido, B., Monterroso, C., Santos-Ucha, P., Prieto-Fernández, Á., 2013. Phytoremediation of hexachlorocyclohexane (HCH)-contaminated soils using Cytisus striatus and bacterial inoculants in soils with distinct organic matter content. Environ. Pollut. 178, 202–210. https://doi.org/10.1016/j.envpol.2013.03.027.

Bell, T.H., Hassan, S.E., Lauron-Moreau, A., Al-Otaibi, F., Hijri, M., Yergeau, E., St-Arnaud, M., 2014. Linkage between bacterial and fungal rhizosphere communities in hydrocarbon-contaminated soils is related to plant phylogeny. ISME J. 8, 331–343. https://doi.org/10.1038/ismej.2013.149.

Benjamini, Y., Hochberg, Y., 1995. Controlling the false discovery rate-a practical and powerful approach to multiple testing. J. R. Stat. Soc. 57, 289–300. https://doi.org/10.2307/2346101.

Bulgarelli, D., Rott, M., Schlaeppi, K., Ver Loren Van Themaat, E., Ahmadinejad, N., Assenza, F., Rauf, P., Huettel, B., Reinhardt, R., Schmelzer, E., Peplies, J., Gloeckner, F.O., Amann, R., Eickhorst, T., Schulze-Lefert, P., 2012. Revealing structure and assembly cues for Arabidopsis root-inhabiting bacterial microbiota. Nature 488, 91–95. https://doi.org/10.1038/nature11336.

Calvelo Pereira, R., Camps-Arbestain, M., Rodríguez Garrido, B., Macías, F., Monterroso, C., 2006. Behaviour of α-, β-, γ-, and δ-hexachlorocyclohexane in the soil–plant system of a contaminated site. Environ. Pollut. 144, 210–217. https://doi.org/10.1016/j.envpol.2005.12.030.

Caporaso, J.G., Kuczynski, J., Stombaugh, J., Bittinger, K., Bushman, F.D., Costello, E.K., Fierer, N., Pena, A.G., Goodrich, J.K., Gordon, J.I., Huttley, G.A., Kelley, S.T., Knights, D., Koenig, J.E., Ley, R.E., Lozupone, C.A., McDonald, D., Muegge, B.D., Pirrung, M., Reeder, J., Sevinsky, J.R., Tumbaugh, P.J., Walters, W.A., Widmann, J., Yatsunenko, T., Zaneveld, J., Knight, R., 2010. QIIME allows analysis of high-throughput community sequencing data. Nat. Methods 7, 335–336. https://doi.org/10.1038/nmeth.f.303.

Chen, Y., Ding, Q., Chao, Y., Wei, X., Wang, S., Qiu, R., 2018. Structural development and assembly patterns of the root-associated microbiomes during phytoremediation. Sci. Total Environ. 644, 1591–1601. https://doi.org/10.1016/j.scitotenv.2018.07.095.

Chu, G., Chen, T., Chen, S., Xu, C., Wang, D., Zhang, X., 2018. The effect of alternate wetting and severe drying irrigation on grain yield and water use efficiency ofIndica-japonica hybrid rice (Oryza sativa L.). Food and Energy Security 7, e133.

Colmer, T.D., 2003. Long-distance transport of gases in plants: a perspective on internal aeration and radial oxygen loss from roots. Plant Cell Environ. 26, 17–36. https://doi.org/10.1046/j.1365-3040.2003.00846.x.

Cuozzo, S.A., Rollán, G.G., Abate, C.M., Amoroso, M.J., 2009. Specific dechlorinase

activity in lindane degradation by Streptomyces sp. M7. World J. Microbiol. Biotechnol. 25, 1539–1546. https://doi.org/10.1007/s11274-009-0039-x.

Cuozzo, S.A., Fuentes, M.S., Bourguignon, N., Benimeli, C.S., Amoroso, M.J., 2012. Chlordane biodegradation under aerobic conditions by indigenous Streptomyces strains. Int. Biodeterior. Biodegradation 66, 19–24. https://doi.org/10.1016/j.ibiod.2011.09.011.

Dai, Z., Hu, J., Xu, X., Zhang, L., Brookes, P.C., He, Y., Xu, J., 2016. Sensitive responders among bacterial and fungal microbiome to pyrogenic organic matter (biochar) addition differed greatly between rhizosphere and bulk soils. Sci. Rep. 6. https://doi.org/10.1038/srep36101.

Dallinger, A., Horn, M.A., 2014. Agricultural soil and drilosphere as reservoirs of new and unusual assimilators of 2,4-dichlorophenol carbon. Environ. Microbiol. 16, 84–100. https://doi.org/10.1111/1462-2920.12209.

Edwards, J., Johnson, C., Santos-Medellín, C., Lurie, E., Podishetty, N.K., Bhatnagar, S., Eisen, J.A., Sundaresan, V., 2015. Structure, variation, and assembly of the root-associated microbiomes of rice. Proc. Natl. Acad. Sci. 112, E911–E920. https://doi.org/10.1073/pnas.1414592112.

Feng, X., Li, J., Teng, Y., Wang, J., 2011. Residues and health risk assessment of organochloring pesticides in soils on the shore of Songhua River in Jilin city, China. Environ. Chem. 30 (9), 1604–1610.

Hayat, T., Ding, N., Ma, B., He, Y., Shi, J., Xu, J., 2011. Dissipation of pentachlorophenol in the aerobic–anaerobic interfaces established by the rhizosphere of rice (L.) root. J. Environ. Qual. 40 (6), 1722–1729. https://doi.org/10.2134/jeq2010.0347.

He, Y., Xu, J., Tang, C., Wu, Y., 2005. Facilitation of pentachlorophenol degradation in the rhizosphere of ryegrass (Lolium perenne L.). Soil Biol. Biochem. 37, 2017–2024. https://doi.org/10.1016/j.soilbio.2005.03.002.

He, Y., Li, X., Shen, X., Jiang, Q., Chen, J., Shi, J., Tang, X., Xu, J., 2015. Plant-assisted rhizoremediation of decabromodiphenyl ether for e-waste recycling area soil of Taizhou, China. Environ. Sci. Pollut. Res. 22, 9976–9988. https://doi.org/10.1007/s11356-015-4179-2.

Heidelberg, J.F., Paulsen, I.T., Nelson, K.E., et al., 2002. Genome sequence of the dissimilatory metal ion–reducing bacterium Shewanella oneidensis. Nat. Biotechnol. 20 (11), 1118–1123.

Ishag, A.E.S.A., Abdelbagi, A.O., Hammad, A.M.A., Elsheikh, E.A.E., Elsaid, O.E., Hur, J., 2017. Biodegradation of endosulfan and pendimethalin by three strains of bacteria isolated from pesticides-polluted soils in the Sudan. Applied Biological Chemistry 60, 287–297. https://doi.org/10.1007/s13765-017-0281-0.

Kidd, P.S., Prieto-Fernández, A., Monterroso, C., Acea, M.J., 2008. Rhizosphere microbial community and hexachlorocyclohexane degradative potential in contrasting plant species. Plant Soil 302, 233–247. https://doi.org/10.1007/s11104-007-9475-2.

Li, Y., Wang, X., 2013. Root-induced changes in radial oxygen loss, rhizosphere oxygen profile, and nitrification of two rice cultivars in Chinese red soil regions. Plant Soil 365, 115–126. https://doi.org/10.1007/s11104-012-1378-1.

Lin, Z., Zhen, Z., Wu, Z., Yang, J., Zhong, L., Hu, H., Luo, C., Bai, J., Li, Y., Zhang, D., 2016. The impact on the soil microbial community and enzyme activity of two earthworm species during the bioremediation of pentachlorophenol-contaminated soils. J. Hazard. Mater. 301, 35–45. https://doi.org/10.1016/j.jhazmat.2015.08.034.

Lin, J., Meng, J., He, Y., Xu, J., Chen, Z., Brookes, P.C., 2018. The effects of different types of crop straw on the transformation of pentachlorophenol in flooded paddy soil. Environ. Pollut. 233, 745–754. https://doi.org/10.1016/j.envpol.2017.10.114.

Liu, J., He, X.X., Lin, X.R., Chen, W.C., Zhou, Q.X., Shu, W.S., Huang, L.N., 2015. Ecological effects of combined pollution associated with e-waste recycling on the composition and diversity of soil microbial communities. Environ. Sci. Technol. 49 (11), 6438–6447. https://doi.org/10.1021/es5049804.

López, L., Pozo, C., Rodelas, B., Calvo, C., Juárez, B., Martínez-Toledo, M.V., González-López, J., 2005. Identification of bacteria isolated from an oligotrophic lake with pesticide removal capacities. Ecotoxicology 14, 299–312. https://doi.org/10.1007/s10646-003-6367-y.

Lu, T., Ke, M., Peijnenburg, W., Zhu, Y., Zhang, M., Sun, L., Fu, Z., Qian, H., 2018. Investigation of rhizospheric microbial communities in wheat, barley and two rice varieties at the seedling stage. J. Agric. Food Chem. 66, 2645–2653. https://doi.org/10.1021/acs.jafc.7b06155.

Maharachchikumbura, S.S.N., Hyde, K.D., Jones, E.B.G., et al., 2016. Families of Sordariomycetes. Fungal Divers. 79, 1–317.

Mei, X.Q., Ye, Z.H., Wong, M.H., 2009. The relationship of root porosity and radial oxygen loss on arsenic tolerance and uptake in rice grains and straw. Environ. Pollut. 157, 2550–2557. https://doi.org/10.1016/j.envpol.2009.02.037.

Mitter, E., Freitas, J., Germida, J., 2017. Bacterial root microbiome of plants growing in oil sands reclamation covers. Front. Microbiol. 8, 849 (doi:10.3389.fmicb.2017.00849).

Mitton, F.M., Gonzalez, M., Monserrat, J.M., Miglioranza, K.S.B., 2016. Potential use of edible crops in the phytoremediation of endosulfan residues in soil. Chemosphere 148, 300–306. https://doi.org/10.1016/j.chemosphere.2016.01.028.

Peiffer, J.A., Spor, A., Koren, O., Jin, Z., Tringe, S.G., Dangl, J.L., et al., 2013. Diversity and heritability of the maize rhizosphere microbiome under field conditions. Proc. Natl. Acad. Sci. 110, 6548–6553. https://doi.org/10.1073/pnas.1302837110.

Peperanney, C., Campbell, A.N., Koechli, C.N., Berthrong, S., Buckley, D.H., 2016. Unearthing the ecology of soil microorganisms using a high resolution DNA-SIP approach to explore cellulose and xylose metabolism in soil. Front. Microbiol. 7, 703. https://doi.org/10.3389/fmicb.2016.00703.

Phillips, T.M., Seech, A.G., Lee, H., Trevors, J.T., 2005. Biodegradation of hexachlorocyclohexane (HCH) by microorganisms. Biodegradation 16, 363–392. https://doi.org/10.1007/s10532-004-2413-6.

Popp, P., Keil, P., Moder, M., Paschke, A., Thuss, U., 1997. Application of accelerated solvent extraction followed by gas chromatography, high-performance liquid chromatography and gas chromatography mass spectrometry for the determination of

polycyclic aromatic hydrocarbons, chlorinated pesticides and polychlorinated dibenzo-p-dioxins and dibenzofurans in solid wastes. J. Chromatogr. A 774, 203–211. https://doi.org/10.1016/s0021-9673(97)00337-3.

Rani, M., Shanker, U., Jassal, V., 2017. Recent strategies for removal and degradation of persistent & toxic organochlorine pesticides using nanoparticles: a review. J. Environ. Manag. 190, 208–222. https://doi.org/10.1016/j.jenvman.2016.12.068.

Robinson, M.D., McCarthy, D.J., Smyth, G.K., 2010. edgeR: a bioconductor package for differential expression analysis of digital gene expression data. Bioinformatics 26, 139–140. https://doi.org/10.1093/bioinformatics/btp616.

Saez, J.M., Álvarez, A., Benimeli, C.S., Amoroso, M.J., 2014. Enhanced lindane removal from soil slurry by immobilized Streptomyces consortium. Int. Biodeterior. Biodegradation 93, 63–69. https://doi.org/10.1016/j.ibiod.2014.05.013.

Sagarkar, S., Mukherjee, S., Nousiainen, A., Björklöf, K., Purohit, H.J., Jørgensen, K.S., Kapley, A., 2013. Monitoring bioremediation of atrazine in soil microcosms using molecular tools. Environ. Pollut. 172, 108–115. https://doi.org/10.1016/j.envpol.2012.07.048.

Salam, J.A., Hatha, M.A.A., Das, N., 2017. Microbial-enhanced lindane removal by sugarcane (Saccharum officinarum) in doped soil-applications in phytoremediation and bioaugmentation. J. Environ. Manag. 193, 394–399. https://doi.org/10.1016/j.jenvman.2017.02.006.

Santos-Medellín, C., Edwards, J., Liechty, Z., Bao, N., Sundaresan, V., 2017. Drought stress results in a compartment-specific restructuring of the rice root-associated microbiomes. mBio 8 (4). https://doi.org/10.1128/mBio.00764-17. (e00764-17).

Shelton, D.R., Khader, S., Karns, J.S., Pogell, B.M., 1996. Metabolism of twelve herbicides by Streptomyces. Biodegradation 7, 129–136. https://doi.org/10.1007/BF00114625.

Shenton, M., Iwamoto, C., Kurata, N., Ikeo, K., 2016. Effect of wild and cultivated rice genotypes on rhizosphere bacterial community composition. Rice 9. https://doi.org/10.1186/s12284-016-0111-8.

Su, Y., Zhu, Y., 2007. Transport mechanisms for the uptake of organic compounds by rice (Oryza sativa) roots. Environ. Pollut. 148, 94–100. https://doi.org/10.1016/j.envpol.2006.11.004.

Sun, X., Zhu, L., Wang, J., Wang, J., Su, B., Du, Z., Guo, P., 2017. Effects of endosulfan on the populations of cultivable microorganisms and the diversity of bacterial community structure in brunisolic soil. Water Air Soil Pollut. 228. https://doi.org/10.1007/s11270-017-3357-x.

Walters, W.A., Zhao, J., Nicholas, Y., Wallace, J.G., Jessica, S., Wei, Z., et al., 2018. Large-scale replicated field study of maize rhizosphere identifies heritable microbes. Proc. Natl. Acad. Sci. 115, 7368–7373. https://doi.org/10.1073/pnas.1800918115.

Wang, M.Y., Chen, A.K., Wong, M.H., Qiu, R.L., Cheng, H., Ye, Z.H., 2011. Cadmium accumulation in and tolerance of rice (Oryza sativa L.) varieties with different rates of radial oxygen loss. Environ. Pollut. 159, 1730–1736. https://doi.org/10.1016/j.envpol.2011.02.025.

Wei, H., Meng, T., Li, C., Xu, K., Huo, Z., Wei, H., Guo, B., Zhang, H., Dai, Q., 2017. Comparisons of grain yield and nutrient accumulation and translocation in high-yielding japonica/indica hybrids, indica hybrids, and japonica conventional varieties. Field Crop Res. 204, 101–109. https://doi.org/10.1016/j.fcr.2017.01.001.

Wick, L.Y., Furuno, S., Harms, H., 2010. Fungi as transport vectors for contaminants and contaminant-degrading bacteria. In: Timmis, K.N. (Ed.), Handbook of Hydrocarbon and Lipid Microbiology. Springer, Berlin, Heidelberg, pp. 1555–1561.

Xu, Y., He, Y., Zhang, Q., Xu, J., Crowley, D., 2015. Coupling between pentachlorophenol dechlorination and soil redox as revealed by stable carbon isotope, microbial community structure, and biogeochemical data. Environ. Sci. Technol. 49, 5425–5433. https://doi.org/10.1021/es505040c.

Xu, Y., Xue, L., Ye, Q., Franks, A., Zhu, M., Feng, X., Xu, J., He, Y., 2018. Inhibitory effects of sulfate and nitrate reduction on reductive dechlorination of PCP in a flooded paddy soil. Front. Microbiol. 9, 567. https://doi.org/10.3389/fmicb.2018.00567.

Xue, L., Feng, X., Xu, Y., Li, X., Zhu, M., Xu, J., He, Y., 2017. The dechlorination of pentachlorophenol under a sulfate and iron reduction co-occurring anaerobic environment. Chemosphere 182, 166–173. https://doi.org/10.1016/j.chemosphere.2017.04.124.

Yu, H., Feng, C., Liu, X., Yi, X., Ren, Y., Wei, C., 2016. Enhanced anaerobic dechlorination of polychlorinated biphenyl in sediments by bioanode stimulation. Environ. Pollut. 211, 81–89. https://doi.org/10.1016/j.envpol.2015.12.039.

Zhang, L., Yang, Q., Wang, S., Li, W., Jiang, S., Liu, Y., 2017. Influence of silicon treatment on antimony uptake and translocation in rice genotypes with different radial oxygen loss. Ecotoxicol. Environ. Saf. 144, 572–577. https://doi.org/10.1016/j.ecoenv.2017.06.076.

Zhang, J.Y., Liu, Y.X., Zhang, N., Hu, B., Jin, T., et al., 2019. NRT1.1B is associated with root microbiota composition and nitrogen use in field-grown rice. Nat. Biotechnol. https://doi.org/10.1038/s41587-019-0104-4.

Zhao, J., Ye, Z., Zhong, H., 2018. Rice root exudates affect microbial methylmercury production in paddy soils. Environ. Pollut. 242, 1921–1929. https://doi.org/10.1016/j.envpol.2018.07.072.

Zhu, M., Zhang, L., Zheng, L., Zhuo, Y., Xu, J., He, Y., 2018. Typical soil redox processes in pentachlorophenol polluted soil following biochar addition. Front. Microbiol. 9, 579. https://doi.org/10.3389/fmic.2018005.